

Docket No.: NEL-006

AMENDMENTS TO THE CLAIMS

Please cancel claim 16 without prejudice or disclaimer to its underlying subject matter. Please amend claims 10-11, 14-15 and 17 as set forth below:

8. (Previously presented) A deoxyribonucleic acid (DNA) vaccine comprising a liposome-encapsulated plasmid containing a gene encoding for hemagglutinin protein.

- 9. (new) The vaccine as claimed in Claim 8 deliverable to a respiratory tract using intranasal administration and/or by aerosol inhalation.
- 10. (currently amended) The deoxyribonucleic acid (DNA) vaccine of claim 8. Use of the vaccine of claim 8 for preventing and/or treating wherein the vaccine prevents or treats an influenza virus infection.
- 11. (currently amended) The deoxyribonucleic acid (DNA) vaccine of claim 8, Use of the vaccine of claim 8 for eliciting wherein the vaccine elicits long-lasting protective antiviral immune responses against influenza viruses.
- 12. (Previously presented) A plasmid vector construct pCI-HA10 comprising a gene encoding for hemagglutinin protein and capable of expressing said hemagglutinin protein in a host.
- 13. (Previously presented) A method for constructing a plasmid pCI-HA10 comprising the following steps:
 - (1) amplifying hemagglutinin gene from viral and mRNA with PCR;
 - (2) inserting and ligating the hemagglutinin gene into a pCI vector;
 - (3) transforming the resulting vector into competent E.coli DH5 α cells;
 - (4) transcribing and translating of pCI-HA10; and
 - (5) preparing and purifying pCI-HA10 by bulk preparation method.



14. (currently amended) A liposome formulation for encapsulating the The method of constructing the plasmid pCI-HA10 of claim 1213, further comprising the step of encapsulating the plasmid pCI-HA10 in a liposome formulation, said liposome formulation comprising of 7% 1,2 dioleoyl-3-dimethylammonium chloride (DODAC), 78% 1,2-dioleoyl-sn-glycrerol-3-phospho-ethanolamine (DOPE) and 15% polyethylene glycol C8 ceramide (PEG₂₀₀₀C₈CER).

- 15. (currently amended) A method for encapsulating The method of constructing the plasmid pCI-HA10 of claim 1213, further comprising the step of encapsulating the plasmid pCI-HA10 in a liposome formulation, into liposomes comprising the following steps:
- (1) preparing 7% DODAC, 78% DOPE, and 15% PEG₂₀₀₀C₈CER at 10mg/ml concentrations to form a lipid film at 50 °C for 2h under vacuum;
 - (2) incubating the lipid film at 50 °C for 2h under vacuum;
- (3) reconstituting the lipid film with distilled water and 1M β -octylglucanopyranoside detergent at 20% of the total preparation volume;
- (4) adding the plasmid DNA to the lipid film at a concentration of 400 μ g DNA/ml of 10 mg/ml;
- (5) transferring the reconstituted preparation into dialysis tubing and dialyzing in 1X HEPES buffer solution (150 mM NaCI, 20 mM Hepes, pH 7.4) at 23 °C for 15 h; and
- (6) removing the free, non-encapsulate DNA from encapsulated DNA on a DEAE Sepharose CL-6B anion exchange column.

16. (cancelled)

- 17. (currently amended) The method of constructing A method of delivering the liposome encapsulated the plasmid pCI-HA10 of claim 16-15, further comprising the step of delivering the liposome-encapsulated pCI-HA10 to a respiratory tract using intranasal administration, and/or aerosol inhalation to eliciting protective antiviral immune responses to influenza viruses.
- 18. (Previously presented) A method for preventing and/or treating influenza virus infection, comprising administering to a patient in need thereof a pharmaceutically effective amount of the vaccine of claim 8.

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19. (Previously presented) A method for eliciting long-lasting protective antiviral immune responses against influenza viruses, comprising administering to a patient in need thereof a pharmaceutically effective amount of the vaccine of claim 8.